

ARTICLE / INVESTIGACIÓN

Effect of D-Aspartic acid on the level of some sex hormones and the biochemical parameters of the blood of Shami Bucks

Ali Shehab Ahmad^{1*}, Safaa Sabbar Atiyah²

DOI. 10.21931/RB/2022.07.04.36

¹Department of Animal Production, College of Agriculture, University of Diyala, Iraq.²Department of Animal Production, Technical Institute of Kufa, Al-Furat Al-Awsat Technical University, Iraq.Corresponding author: dr.alishehab@gmail.com

Abstract: This study was conducted at the Animal Production Department/ College of the Agriculture/ University of Diyala from 15/9/2021 to 15/10/2021 to investigate the effect of injecting D-aspartic amino acid in Shami Bucks on some blood biochemical and hormonal characteristics. Twelve's Shami Bucks aged between 1.5-2 years, and body weight ranged between 35-40 kg. The animals were divided into four groups (treatments) with three replicates among each group as follows, T₁ (control group) was injected with normal saline only, T₂, T₃ and T₄ groups were injected i.m. with D-aspartic acid as follows, 125 mg, 250 mg and 375 mg for T₂, T₃ and T₄ groups respectively, every 48 hours in the afternoon. Blood samples were collected from the jugular vein, and serum was taken and stored at -20 ° C until analyzed. The results of the present study also indicated significant differences (P<0.05) of FSH (1.37±2.59, 1.45±0.89, 1.87±1.76 and 0.77±0.45) and LH (1.96±1.56, 2.19 ± 0.22, 2.22±1.44 and 1.11±1.30) respectively for the T₂, T₃ and T₄ treatments as compared with the T₁ (control group). The results showed a significant increase (P<0.05) of total protein (6.23±0.02, 6.26±0.39, 6.46±1.23 and 4.35±0.12), albumin (4.36±1.24, 4.56±1.00, 4.75±1.34 and 3.34±0.11), globulin (1.87±1.33, 1.70±0.11, 1.71±0.01 and 1.01±1.22) and blood urea (6.45±0.23, 6.43±1.39, 6.56±1.56 and 5.22±1.25) respectively for the T₂, T₃ and T₄ treatments as compared with the T₁ (control group). While no significant differences between all experimental treatments in the concentrations of thyroid hormones (T₄, T₃), cholesterol and triglycerides. It can be concluded from the present study that injection of D-aspartic acid had a significant effect on some biochemical blood traits and the level of pituitary sex hormones.

Key words: D-Aspartic acid, Shami Bucks, biochemical parameters, sex hormones.

Introduction

Goats are considered one of the critical economic animals in Iraq, as they are raised for meat, milk, skin and hair, but they live outside pastoralism and agriculture and feed on waste and jungles. Their fertility and productivity are low, so they need to be cared for and their fertility improved¹. The Shami goat originated in the Sham² and was introduced to Cyprus from Syria more than 70 years ago to improve the performance of their goats. Thus, it was called the Cypriot goat. This type of goat was introduced to Iraq (Ruminant Research Station/ General Authority for Agricultural Research/ Ministry of Agriculture) in the name of the Cypriot Shami goats after they were imported from Cyprus through the Ministry of Agriculture in 2006³. Several studies have indicated that environmental factors significantly affect the reproductive efficiency of farm animals, such as increasing the daily light duration and high temperatures⁴⁻⁵. The measurement of hematological and biochemical parameters is essential in evaluating animal activity. It indicates animal health, reflected in the production characteristics, as the blood and biochemical parameters are affected by breed, age, physiological condition, season, nutrition, etc. The animal body's physiological regulation works well, which is reflected in its ability to adapt to circumstances and prevailing environmental conditions⁶⁻⁸. (9) indicated that amino acids such as arginine and aspartic affect the circulatory system by increasing the heart rate without affecting the number

of heartbeats and increasing blood flow to the blood vessels, leading to increased blood circulation. Because there is no study in Iraq to our knowledge showing the effect of injecting aspartic acid on some biochemical parameters of Shami Bucks. This study was conducted to find out the impact of injecting the amino acid L-aspartic by measuring the following characteristics, Total protein, Albumin, Globulin, blood urea, Cholesterol, Triglycerides, Follicle Stimulating Hormone (FSH), Luteinizing Hormone (LH), T₃ (Tri-iodothyronine) and T₄ (Thyroxine).

Materials and methods

This study was conducted at the Animal Production Department/ College of the Agriculture/University of Diyala from 15/9/2021 to 15/10/2021. In the present experiment, twelve Shami Bucks of 1.5-2 years of age and 35-40 kg weight, placed inside semi-open pens in individual cages, were used. The animals were fed with the 1 kg concentrate feed, in addition to giving them hay and green fodder, according to the animal's need (in an open manner) with the availability of drinking water throughout the experiment. The animals were divided into four treated groups with three replicates, T₁ (control group) was injected with normal saline i.m., T₂, T₃ and T₄ were injected with aspartic acid with a concentration

Citation: Ahmad, Ali.; Atiyah, Safaa. Effect of D-Aspartic acid on the level of some sex hormones and the biochemical parameters of the blood of Shami Bucks. *Revis Bionat* a 2022;7(4) 36. <http://dx.doi.org/10.21931/RB/2022.07.04.36>

Received: 25 August 2022 / **Accepted:** 12 October 2022 / **Published:** 15 November 2022

Publisher's Note: Bionatura stays neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Copyright: © 2022 by the authors. Submitted for possible open access publication under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).



of 125 mg, 250 mg and 375 mg i.m. respectively every 48 hours in the afternoon. At the end of the experiment, blood samples were collected from the jugular vein using sterile 8ml syringes with a test tube free of anticoagulant, centrifuged at a speed of 3000 rpm for 15 minutes. Serum was withdrawn using a special pipette, numbered and kept in the freezer at -20 °C until all the analyzes of the biochemical components were completed.

Total protein

The measurement depends on the occurrence of a color change due to the interaction of copper in a base solution of the measuring kit (prepared by the German Human company) with the peptide bonds of the protein that is found in the blood serum. The solution is mixed and left at room temperature for 10 minutes; a spectrophotometer extracts it at a wavelength of (520) nm. The following equation is applied to obtain the total protein level¹⁰.

Blood urea

The hydrolysis of urea to ammonium ions and carbon dioxide is the basis for measuring urea in the blood, which was approved by the Human German company manufacture of urea measurement kits. Ammonium ions with chlorine and salicylate produce a blue-green complex. This complex and its concentration are measured by a spectrophotometer on A wavelength (600), which indicates the amount of urea in the blood.

Cholesterol

The level of cholesterol was measured in the blood serum using a spectrophotometer with a wavelength of (500) nm using ready solutions (Kit)¹¹.

Triglycerides

The triglyceride concentration was estimated using a measuring kit supplied by the Human German company, based on the method of (12), and by a spectrophotometer at a wavelength of (500) nm.

Hormonal assays

The levels of T₃, T₄, FSH, and LH hormones were measured using an Enzyme-linked immunosorbent assay (ELISA) and a kit to measure each hormone supplied by Novormon company.

Statistical Analysis

The experiment was carried out using a completely random design (CRD), and data were analyzed using the statistical program SPSS (SPSS, 2011); the averages of the coefficients for each treatment were compared using Duncan's polynomial test¹³ to determine the significance of differences between the means. According to the following mathematical model: $Y_{ij} = \mu + \tau_i + e_{ij}$.

Results and discussion

The results of the present study indicated a significant ($P < 0.05$) effect of FSH and LH hormones for T₂ (125 mg), T₃ (250 mg) and T₄ (375 mg) compared to T₁ (control group), which were 1.37 ± 2.59 , 1.45 ± 0.89 , 1.87 ± 1.76 and 0.77 ± 0.45 for FSH and 1.96 ± 1.56 , 2.19 ± 0.22 , 2.22 ± 1.44 and 1.11 ± 1.30 for LH respectively (Table 1). There were no significant differences between all treatments of the experiment and the control group in thyroid hormone concen-

trations (T₄, T₃) and the control group (Table 1). D'Aniello *et al.*¹⁴ indicated that the level of aspartic acid was elevated in the hypothalamus, pituitary, and blood serum, which reached its highest level after one hour of injection. When comparing them, the highest concentration was found in the hypothalamic glands for the presence of enzymes that convert L-Asp to D-Asp in the gland tissue. These results were also confirmed by Di Fiore *et al.*¹⁵ of the increase in the activity of Neuro-steroidogenic enzymes in the brains of mice injected with aspartic acid.

Sheffield *et al.*¹⁶ found a decrease in the concentration of aspartic acid in sheep's blood plasma and body tissues fed a low concentration of aspartic acid. Still, if fed a diet rich in aspartic acid, the concentration will increase in both body tissues, the brain and endocrine glands. Boni *et al.*¹⁷ showed a high percentage of aspartic acid in the tissues of both the pineal gland and the pituitary gland. Still, the pituitary gland was the most efficient in storing aspartic acid in sheep. It recorded that the concentration of aspartic acid tripled more than the average concentration in the pituitary gland, ovary, brain and blood serum after 12 hours of aspartic acid treatment. There are more than 700 amino acids, but only 20, including aspartic acid, are the building units of proteins in cells¹⁸.

The results showed a significant increase ($P < 0.05$) of total protein, albumin, globulin and blood urea for the T₂ (125 mg), T₃ (250 mg) and T₄ (375 mg) treatments compared with the T₁ (control group) (table 2). While no significant differences among the treatments group (T₂, T₃ and T₄). The results also showed no significant differences among all experiment treatments and control groups in the concentrations of cholesterol and triglycerides (Table 2).

Amino acids are of great physiological importance, serving as building units for proteins and substrates to synthesize low-molecular-weight substances. Depending on growth or nitrogen balance, amino acids have traditionally been classified as nutritionally essential or non-essential for animals¹⁹. Abdelhamed *et al.*²⁰ mentioned that the concentration of aspartate was the lowest in the local goat breed, while it was the highest in the hybrid goat breed, and this may be due to the breed effect on amino acid production.

Conclusions

The present study showed that injection of D-aspartic acid significantly affected some sex hormones and the biochemical parameters of the blood of local Shami Bucks.

Funding

Self-funding.

Conflicts of Interest

There is no conflict.

Bibliographic references

1. Amasha, M. Gh., Al-Baraka, F. S. and Al-Hamid, S. M. Breeding and caring for local and Shami goats. National Center for Agricultural Research and Technology Transfer. The Hashemite Kingdom of Jordan, 2003.
2. Al-Kass, J. E., Al-Jalili, Z. F. and Aziz, D. E. Sheep and goat principles of production and breeding. College of Agriculture, University of Baghdad. National Library Printing and Publishing Press (Baghdad/ Iraq), 1993, PP. 163- 164.

Hormones concentration	Treatments			
	T ₁ (control)	T ₂ (125 mg)	T ₃ (250 mg)	T ₄ (375 mg)
FSH (ng/ml)	0.77±0.45 b	1.37±2.59 a	1.45±0.89 a	1.87±1.76 a
LH (ng/ml)	1.11±1.30 b	1.96±1.56 a	2.19±0.22 a	2.22±1.44 a
Thyroxin (T₄) (ng/ml)	61.38±1.23 a	61.59±1.2 a	61.55±0.55 a	63.66±0.94 a
Triiodothyronine (T₃) (ng/ml)	2.08±1.16 a	2.47±0.44 a	2.49±1.66 a	2.43±1.70 ¹ a

¹Means with different letters within one row indicate significant differences (p < 0.05).

Table 1. Effect of aspartic acid injection on thyroid and some pituitary hormones of Shami Bucks (mean ± standard error).

Blood Parameters	Treatments			
	T ₁ (control)	T ₂ (125 mg)	T ₃ (250 mg)	T ₄ (375 mg)
Total protein g/dl	4.35±0.12 b	6.23±0.02 a	6.26±0.39 a	6.46±1.23 a
Albumin g/dl	3.34±0.11 b	4.36±1.24 a	4.56±1.00 a	4.75±1.34 a
Globulin g/dl	1.01±1.22 b	1.87±1.33 a	1.70±0.11 a	1.71±0.01 a
Blood urea mg/dl	5.22±1.25 b	6.45±0.23 a	6.43±1.39 a	6.56±1.56 a
Cholesterol mg/dl	19.99±2.22 a	21.11±1.00 a	20.48±1.33 a	21.44±0.76 a
Triglyceride mg/dl	89.88±0.76 a	91.32±2.15 a	90.79±1.00 a	90.76±1.44 ¹ a

Table 2. Effect of aspartic acid injection on some blood biochemical traits of Shami Bucks (mean ± standard error).

- Al-Amri, M. H. M. Effect of treatment with Kisspeptin hormone, GnRH and hCG on reproductive performance of male and female Cypriot goats outside the reproductive season. PhD thesis. College of Agriculture, University of Baghdad. 2015.
- Walkden-Brown, S. W., Restall, B. J. and Henniawati. The male effect in the Australian cashmere goat. Enhancement with buck nutrition and use of oestrous females. Anim. Reprod. Sci., 1993, 32 (1-2): 69- 84.
- Khalil, R. I. Effect of Autumn and Winter seasons on seminal traits and testicular measurements of Awassi ram lambs. Diyala Agric. Sci. J., 2018. 10(2):1-11.
- Hobi, A. A. A study of some blood hematology for different genetic groups of sheep in Iraq. J. Kerbala Univ. Sci., 2012, 10 (2):125-141.
- Al-Rubaie, H. M., Abd, H. A. and Obaid, H. R. Study of the relationship of some metabolic changes with hormonal changes during pregnancy in local Awassi sheep. Kerbala Univ. Sci. J., 2015, 13 (2): 306 - 313.
- Abdel Latif, F. H. Study of some hematological and biochemical parameters and their relationship to milk production in Awassi sheep and local goats. Al-Furat J. Agri. Sci., 2017, 9 (2): 102-134.
- Koifman, B., Wollman, Y., Bogomolny, N., Chernichowsky, T., Finkelstein, A., Peer, G., Scherez, J., Blum, M., Laniado, S., Iaina, A.; Keren, G. Improvement of cardiac performance by intravenous infusion of L-arginine in patients with moderate congestive heart failure. J. Am. Coll. Cardiol. 1995, 26(5):1251-1256.

10. Green, S. A., Jenkins, S. J.; Clark, P. A. A comparison of chemical and electrophoretic methods of serum protein determination in clinically normal domestic animals of various ages. *Cornell Vet.*, 1982, 72 (4):416-426.
11. Allain, C. C., Poon, L. S., Chan, C. S., Richmond, W.; Fu, P. C. Enzymatic determination of total Serum cholesterol. *Clin. Chem.*, 1974, 20(4): 470- 475.
12. Toro, G.; Ackermann, P. G. *Practical clinical chemistry*. Little Brown Company. Boston, Philadelphia, 1975, pp:354.
13. Duncan, D. Multiple Range and Multiple F Tests. *Biometrics*, 1955, 11(1): 1-42.
14. D'Aniello, G., Tolino, A., D'Aniello, A., Errico, F., Fisher, G. H.; Di Fiore, M. M. The role of D-aspartic acid and N-methyl-D-aspartic acid in the regulation of prolactin release. *Endocrinology*, 2000, 141(10): 3862-3870.
15. Di Fiore, M. M., Santillo, A., Falvo, S., Baccari, G. C., Venditti, M., Russo, F. D. G., Lispi, M. and D'Aniello, A. Sex hormone levels in the brain of d-aspartate-treated rats. *C. R. Biol.*, 2018, 341(1): 9-15.
16. Sheffield, J., Roman, C., Roper, B. L., Poole, R. K.; Pickworth, C. L. Flushing and Synchronization Protocol Impacts on out of Season Breeding in Ewes. *J. Anim. Sci.*, 2018, 96 (1): 76.
17. Boni, R., Santillo, R., Macchia, G., Spinelli, P., Ferrandino, G.; D'Aniello, A. D-Aspartate and reproductive activity in sheep. *Theriogenology*, 2006, 65(7): 1265-1278.
18. Wu, G. *Amino Acids: Biochemistry and Nutrition* (1st ed.), CRC Press, Boca Raton, 2010, <https://doi.org/10.1201/b14661>.
19. Wu, G., Bazer, F. W., Dai, Z., Li, D., Wang, J.; Wu, Z. *Amino Acid Nutrition in Animals: Protein Synthesis and Beyond*. *Annu. Rev. Anim. Biosci.*, 2014, 2: 387- 417.
20. Abdelhamed, W. A., El-Danasoury, M. M., El-Hamamsy, S. M.; El-Sayed, M. A. Amino acids profile of three different Egyptian goat breeds. *Egypt. J. of Appl. Sci.*, 2020, 35 (12): 261- 271.